The Development of European Sea Bass (Dicentrarchus labrax L., 1758) Eggs in Relation to Temperature

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Abstract: In this study, the development of European Sea Bass (Dicentrarchus labrax L., 1758) eggs was investigated at different temperatures. The experiments were carried out at 15°C and 17°C. The average diameters of the eggs were determined tobe approximately 1.162±0.004 mm. The embryonic development of the eggs was observed every hour and then photographed. The hatching rates were found to be 84.8-89.4% at 15°C and 82.3-86.3% at 17°C, respectively. Hatching of the larvae took 87 hours at 15°C and 69 hours at 17°C.

Key Words: Sea bass, egg, temperature, embryology

Sıcaklığın Levrek (Dicentrarchus labrax L., 1758) Yumurtalarının Gelişimi ile İlişkisi

Özet: Bu çalışmada, iki farklı sıcaklıkta levrek yumurtalarının gelişimleri incelenmiştir. Denemeler 15° ve 17°C'de olmak üzere iki değişik sıcaklıkta yapılmıştır. Yumurtaların ortalama çapları 1.162±0.004 mm. olarak tespit edilmiştir. Yumurtalardaki embriyolojik gelişim her saatte bir gözlenerek fotoğrafları çekilmiştir. Açılım oranları 15°C'de %84.8-89.4 ve 17°C'de %82.3-86.3 olarak ispat edilmiştir. Larvalar 15°C'de 87 saatte ve 17°C'de 69 saatte yumurtadan çıkmışlardır.

Anahtar Sözcükler: Levrek, yumurta, sıcaklık, embriyoloji

Introduction

The increasing market value of the sea bass and its appropriacy to mass production have led to increasing interest in this species especially in Mediterranean countries. Since the 1970's a number of studies have been carried out on its biology, larvae production and breeding (1-4).

Various studies have also been conducted on the properties and the development of sea bass eggs. It was reported that the size of the egg increased the survival rate and that the eggs hatched by means of hormones were smaller in size (5, 6). Feeding of the adults under favourable conditions affects the quality and the quantity of the eggs positively (7). It was also reported that an increase of 2-3‰ in salinity during incubation would be risky (8).

The main factor affecting the rate and the quality of the embryonic development is temperature. Going

beyond optimum limits during incubation leads to the deterioration of the cellular symmetry and the breaking of the fat drop; it also causes mass mortality and consequently a drop in the rate of larvae production during gastrulation (9). In related studies, different hatching rates of the larvae have been investigated (2-4, 7). In this study, hatching rates of $15^{\circ}C$ (10, 11) and $17^{\circ}C$ (9, 10), which is accepted as the upper limit of optimum temperature for incubation, were investigated.

Materials and Methods

The eggs were taken from wild breeders. After statistical evaluations, the eggs were stocked in an incubator (mesh size 425μ) as 2500 egg/l. The experiments were carried out at two different temperatures 15° C (group A) and 17° C (group B). Each group consisted of two incubators with a volume of 50 l.

During the experiment, the flow rate into tanks was 0.2 m³ per hour. Experiments took place in complete darkness.

Ten samples were taken from each incubator and photographed every hour to determine the common embryonic developments. Whenever there was an evident difference during the embryonic development, the photographs were taken before or after the due time.

The volumetric method was used to determine the survival rate and stock density of the eggs. From a homogenous mixture of the larvae and the eggs in the incubators five samples were taken with a 10 ml pipette. The average of the nearest three samples was found in order to determine the survival rates of the larvae.

Before the eggs used during the experiment were placed in the incubators, 30 samples were taken, and the mean diameter and their standard deviation calculated. After statistical analyses were done, the eggs were distributed among the incubators. The significance rate for the relations among the groups was obtained by carrying out the significance test for the difference between the two percentages in independent groups within a 95% confidence interval (11). Statistical difference was considered to be significant when the P value was >0.05.

Results

During the incubation, the salinity of natural sea water varied between 36.9 and 37.2‰. Also the oxygen levels varied between 6.4 and 7.2 mg/l. In group A and group B, the changes in temperature levels were 14.7-15.3°C and 16.9-17.3°C, respectively.

In this study, the eggs were taken from wild breeders. The diameter of the average egg was determined to be 1.162 ± 0.004 mm (Fig. 1-A). The statistical assessment of the eggs used in the experiment is shown in Table.1.

Table 1. The average egg diameters (mm) in sea bass.

N X ort. ±Sx		Min	Max.		
30	1.162±0.004	1.088	1.214		

During the incubation (group A) a two-cell stage was observed at 15° C, 1.25h after fertilization. The second division occurred 1.50h after the fertilization and a four-

cell stage was observed. An 8-cell stage appeared after 2.40h. It was hard to observe the other symmetrical divisions, but they continued to divide. Morula and blastula stages were observed after 5.05h and 9.30h, respectively. In group B, eggs were incubated at 17°C; these stages were observed at 1.10h (Fig.1-B), 1.45h (Fig.1-C), 2.30h (Fig.1-E), 4.30h (Fig.1-H) and 8.30h (Fig.1-M), respectively. For group A, the gastrulation started 13.40 h after the fertilization and continued for 28.35h. For group B, it was between 13.00 and 22.55h after the fertilization. The germ disc covered half of the egg in 23.35h and $\frac{3}{4}$ of the egg in 26.50h. Half an hour after this period, the embryo started to become denser before closing blastopore. After 27.20h, the embryo was slightly visible. 38.50h after the fertilization, 5-6 couples of somit were observed. 41h later the accumulation of the oil globule, and 42h later the kupffer apparatus were observed. 42.40h later, the formation of the heart started and 43.50h later, pigmentation started. The embryo surrounded 2/3 of the egg 47.20h later and covered $\frac{3}{4}$ of it 51.30h after the fertilization. 58.50h later, the formation of the optic cup started and 60.30h later the primordial fin started to form. 65.30h after the fertilization the heartbeat started. Then, the number of mesoderm somits increased while the embryo was growing in the egg. The star-shaped pigmentation occurred on the cranium. 84.25h after the fertilization of the 10% of the larvae, and 87.10h later, 100% of them splitted the corion with the help of the enzyme secretion, excreted from the cranium of larvae and released from the egg. In group B, these stages were observed as follows: 18.30h (Fig. 2-G), 20.30h (Fig. 2-J), 21.30h (Fig. 2-K), 29.00h (Fig. 3-D), 29.30h (Fig. 3-E), 30.30h (Fig. 3-F), 33.30h (Fig. 3-J), 35.00h (Fig. 3-L), 35.30h (Fig. 3-M), 44.30h (Fig. 4-F), 49.00h (Fig. 4-L), 50.30h (Fig. 4-M), 52.30h (Fig. 4-P), 53.30h (Fig. 4-R), 64.00h (Fig. 5-L), and 68.00h (Fig. 5-P), respectively. At 15°C, hatching rates were 89.4% for A1 incubator and 84.8% for A2. At 17°C, they were 86.3% for B1 and 82.3% for B2.

The hatching rate of the eggs in group A incubators was found to be 87.1%, and it was 84.3% in group B. When the significance test for the difference between two percentages in independent groups was applied, the freedom degree (SD) was found to be 169 and the standard deviation (sd) 0.053. The t value in 0.05;150 coordinates was examined within 95% confident interval and the difference between the two groups was found to be statistically insignificant (t=0.56, p>0.05).

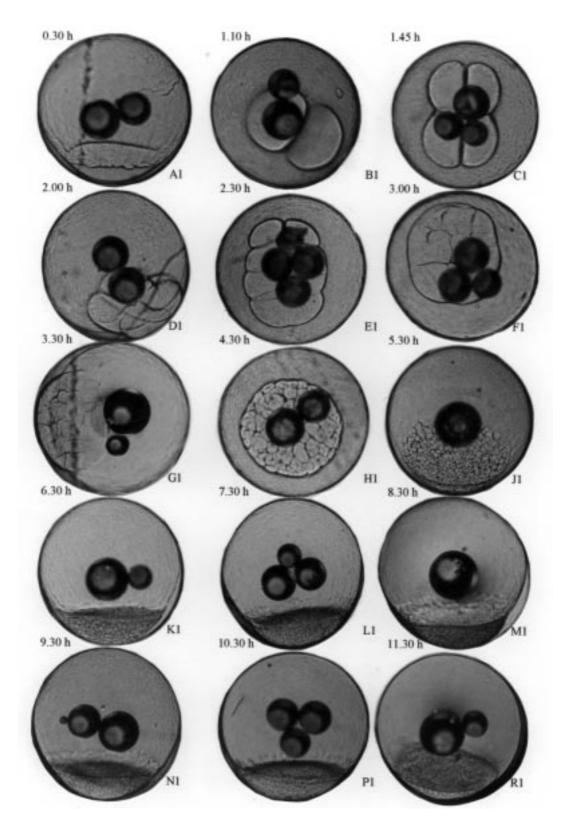


Figure 1. The embryonic development of sea bass (Dicentrarchus labrax L.) eggs between 0:30 and 11:30 hours (Original).

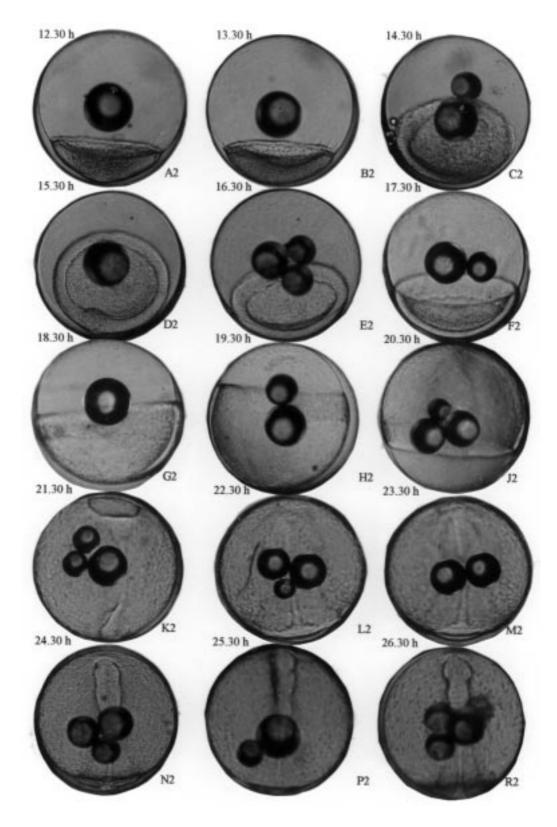


Figure 2. The embryonic development of sea bass (Dicentrarchus labrax L.) eggs between 12:30 and 25:30 hours (Original).

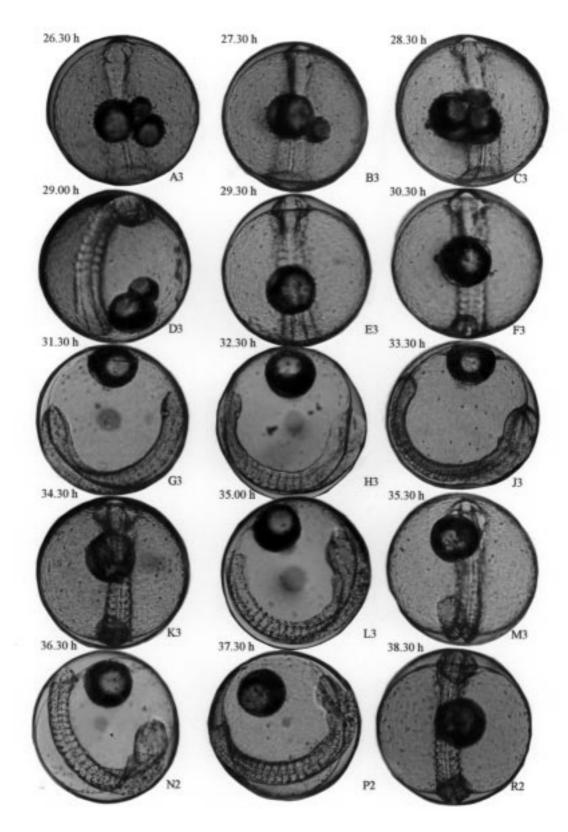


Figure 3. The embryonic development of sea bass (Dicentrarchus labrax L.) eggs between 26:30 and 38:30 hours (Original).

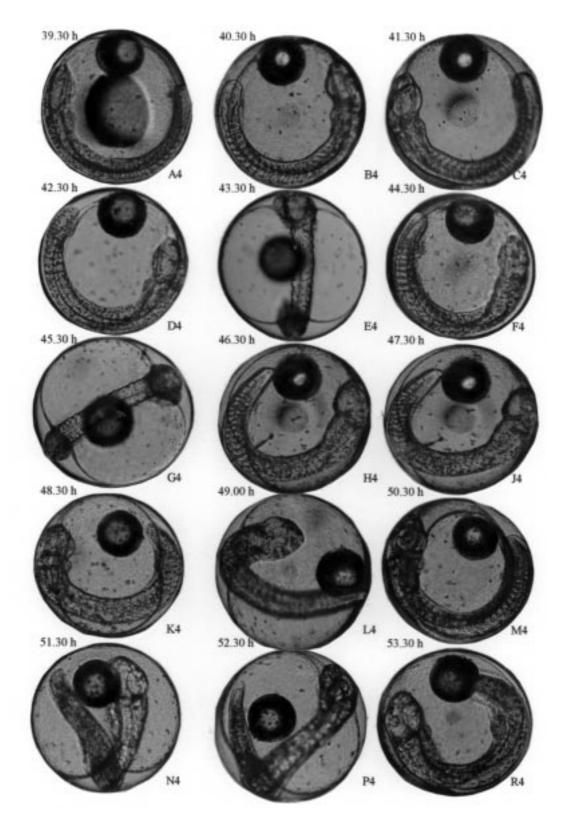


Figure 4. The embryonic development of sea bass (Dicentrarchus labrax L.) eggs between 39:30 and 53:30 hours (Original).

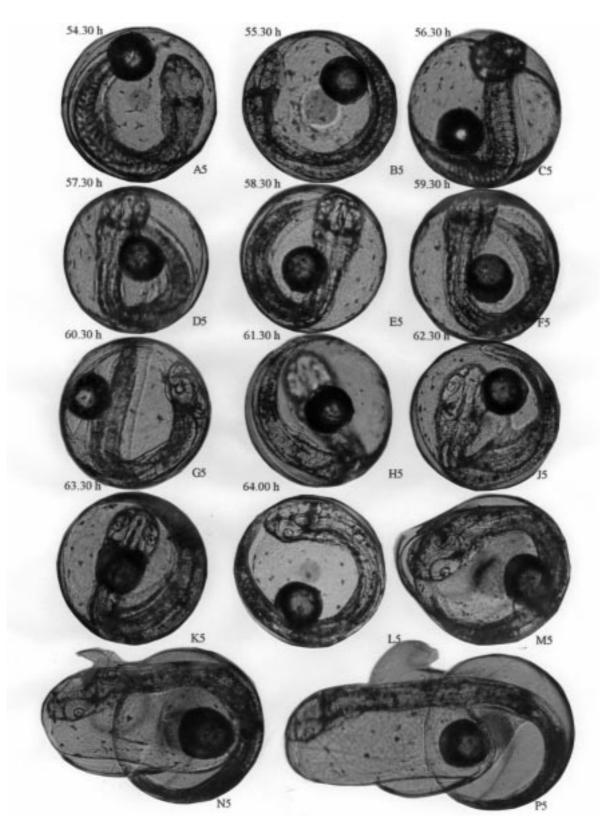


Figure 5. The embryonic development of sea bass (Dicentrarchus labrax L.) eggs between 54:30 and 64:00 hours (Original).

Discussion

The minimum oxygen rate in the incubators was determined to be 6.4 mg/l and maximum 7.2 mg/l. The temperature levels varied for group A between 14.7 and 15.2°C during incubation. For group B, it was 16.8-17.1 mg/l. These values were different from those in previous studies (1-6), which used 13-15°C.

The sea bass exhibiting diadromous properties leaves its eggs in natural marine environments. During incubation studies, the eggs demonstrated pelagic properties in salinities over 35% and demersal properties in salinities below 34% (6, 12). As in these studies, natural sea water was used during incubation. It has been reported that oxygen levels between 5 and 7 mg/l in the incubation of sea bass eggs have affected the embryonic development in a positive way (13, 14). In this study, the oxygen levels in all the incubators was kept within specified limits. Oxygen and salinity differences in this study were kept within the values at which embryonic developments in nature usually occurred, and thus it was attempted to prevent their adverse effects on the incubation of the eggs. When the egg completed its embryonic development, the initial hatching period for 10% was determined to be 84.25h at 15°C and 64.00h at 17°C. 100% hatching occurred after 87.10h at 15°C and after 68.00h at 17°C. (14) reported 87.00h and 93.00h for hatching rates of 10% and 100% respectively. (15) explained that 100% hatching was observed after 110.50h at 13°C, (9) 108.00h at 13.6°C, (16) 104.00h at 15°C, (17) 115.00h at 13°C and (18) 110.00h at 14°C (Table 2).

The incubation temperature initiated premature hatching in about 20 hours at 15° C, and 40 h at 17° C. In spite of this, the embryonic development occurred without any abnormalities. At 15° C the hatching rate was determined to be 89.4% for the A1 incubator, 84.8% for A2, and at 17° C 86.3% for B1 and 82.3% for B2.

In this study, the development of eggs was established at hourly intervals. Therefore, it is important that these photograph series provide certain results to hatcheries taking eggs from different farms, and supply them with information on which period the eggs are at and when they are expected to hatch.

Table 2.	The comparison of embryonic developments. Results (Results of this study), 1*. Salvatorelli et al.(15), 2*. Jennings and Pawson (9), 3*.
	Uçal (16), 4*. Devauchelle and Coves (17), 5*. Barnabe (18), 6*. Marino et al. (14).

	Resources and Experimental Temperatures								
Stages of Embryonic Development	 Results*		1*	2*	3*	4*	5*	6*	
	15°C	17°C	13°C	13.6°C	15°C	13°C	14°C	13°C	
2nd Blastomere	1:25	1:10					1:30		
4th Blastomere	1:50	1:45					2:00		
8th Blastomere	2:40	2:30		16:00	2:15		2:30		
Morula	5:05	4:30				8:20	4:30	7:10	
Blastula	9:30	8:30			8:30				
Starting of Gastrulation	13:40	13:00		26:00	20:00	24:00			
Gastrulation ¹ / ₂	23:35	18:30		36:00	35:00				
Gastrulation $\frac{3}{4}$	26:50	20:30			40:00				
Observation Embryo Profile	27:20	21:30					40:00	31:30	
Gastrulation ⁴ / ₄	28:35	22:55	24:00	48:00					
The Formation of Somit	38:50	29:00			55:00			36:00	
The Accumulation of Oil Globule	41:10	29:30							
The Appearance of Kupffer	42:00	30:30							
The Appearance of Pigmentation	42:40	33:30	72.00		80:00		69.00		
The Appearance of Heart	43:50	35:00			55:00				
Embryo ² /3	47:20	35:30		78:00					
Embryo ³ / ₄	51:30	44:30		86:00	90:00				
The Formation of Optic Cup	58:50	49:00					80:00	40:45	
The Formation Primordial	60:30	50:30					85:00		
The First Heartbeat	65:30	53:30				74:00		62:30	
Hatching (10%)	84:25	64:00						87:00	
Hatching (100%)	87:10	68:00	110:50	108:00	104:00	115:00	110:00	93:00	

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